

In the Claims:

1. (Previously presented) A multilayer device for use in tissue engineering, comprising:

(a) at least a first layer comprised of a polymer scaffold having a pattern of microchannels therein and

(i) wherein the microchannels are suitable for the attachment and culturing of animal cells within the microchannels, and

(ii) wherein the microchannels are connected for the circulation of fluid through the first layer, and

(b) at least a second layer comprised of a polymer scaffold,

wherein the first and second layers are joined or fastened together and the first layer is formed by forming a mold from a substrate material using a photoresist processing technique that includes:

i) coating the substrate material with a photoresist; and ii) forming a pattern in the photoresist, and casting the first layer on the respective mold.

2. (Cancelled)

3. (Cancelled)

4. (Cancelled)

5. (Previously presented) The multilayer device of claim 1, wherein the polymer of the first layer is biodegradable.

6. (Previously presented) The multilayer device of claim 1, wherein the polymers of all the layers of the multilayer device are biodegradable.

7. (Previously presented) The multilayer device of claim 1, wherein the second layer has a pattern of microchannels therein.

8. (Previously presented) The multilayer device of claim 7, wherein the pattern in the first and second layers are similar.

9. (Previously presented) The multilayer device of claim 7, wherein the pattern in the first and second layers are different.

10. (Previously presented) The multilayer device of claim 7, wherein the pattern in the first layer is suitable for the culturing of endothelial cells and the layer in the second layer is suitable for the culturing of parenchymal cells.

11. (Previously presented) The multilayer device of claim 7, wherein the patterns of the first and second layers are aligned to form a vasculature.
12. (Previously presented) The multilayer device of claim 1, wherein the channels of the first layer are connected beginning from one or more inlets, expanding into more channels, and then converging back into one or more outlets.
13. (Previously presented) The multilayer device of claim 1, wherein the channels of all the layers of the multilayer device are connected beginning from one or more inlets, expanding into more channels, and then converging back into one or more outlets.
14. (Previously presented) The multilayer device of claim 1, further comprising a third layer comprised of a micromachined polymer scaffold suitable for attachment and culturing of animal cells, wherein the first, second and third layers are joined or fastened together.
15. (Previously presented) The multilayer device of claim 14,
 - (a) wherein the second layer is unpatterned; and
 - (b) wherein the third layer has a pattern of channels therein,
 - (i) wherein the channels are suitable for the attachment and culturing of animal cells within the channels, and
 - (ii) wherein the channels are connected for the circulation of fluid through the layer.
16. (Previously presented) The multilayer device of claim 15, wherein the pattern in the first and third layers are similar.
17. (Previously presented) The multilayer device of claim 15, wherein the pattern in the first and third layers are different.
18. (Previously presented) The multilayer device of claim 17, wherein the pattern in the first layer is suitable for the culturing of endothelial cells and the layer in the third layer is suitable for the culturing of parenchymal cells.
19. (Previously presented) The multilayer device of claim 1, wherein the animal cells are selected from the group consisting of endothelial cells, parenchymal cells, bone marrow cells, osteoblasts, mesenchymal stem cells, satellite cells, and fibroblasts.
20. (Previously presented) The multilayer device of claim 1, wherein the cells cultured in the channels of the first layer are endothelial cells.

21. (Previously presented) The multilayer device of claim 1, wherein one or more of the layers comprise through-holes.
22. (Previously presented) The multilayer device of claim 1, wherein one or more of the layers comprise an alignment indentation on the surface of a layer and an alignment protrusion on an opposing surface of a layer, the alignment indentations shaped to mate with the alignment protrusion.
23. (Previously presented) The multilayer device of claim 1, wherein the first layer is subdivided into zones of animal cell support.
24. (Previously presented) The multilayer device of claim 23, wherein the zones of animal cell support comprise cell adhesion molecules.
25. (Currently amended) A method of making a multilayer device, comprising the steps of:
- (a) fabricating at least a first layer comprised of a polymer scaffold suitable for attachment and culturing of animal cells and having a pattern of channels therein, wherein the at least a first layer is fabricated by forming a mold from a semiconductor substrate material using a photoresist processing technique that includes manufacturing process coating the semiconductor substrate material with a photoresist, and forming a pattern in the photoresist, and then casting the at least a first layer on the mold, and
 - (i) wherein the channels are suitable for the attachment and culturing of animal cells within the channels,
 - (ii) wherein the channels are connected for the circulation of fluid through the layer, and
 - (iii) wherein at least one of the channels is about 30-200 microns in width and about 200 microns in depth;
 - (b) obtaining at least a second layer for supporting animal cell growth wherein the second layer is comprised of a polymer scaffold suitable for attachment and culturing of animal cells; and
 - (c) fastening together the first and second layers to form lumens of the channels.
26. (Previously presented) The method of claim 25, wherein the joining or fastening is by a method selected from the group consisting of the methods of solvent bonding; reflow by heating; treating the surface of the layer with oxygen plasma; polymer flow at the surface of the layer, mechanically fastening the layers with fasteners selected from the group comprising barbs, pins, screws, clamps, staples, wires, string, and sutures; and adhering the layers by the use of adhesives, adhesive films or adhesive layers.

27. (Cancelled) A method of making a multilayer device, comprising the steps of:
- (a) obtaining a layer comprised of a material suitable for attachment and culturing of animal cells and having a pattern of channels therein,
 - (i) wherein the channels are suitable for the attachment and culturing of animal cells within the channels, and
 - (ii) wherein the channels are connected for the circulation of fluid through the layer; and
 - (b) folding or rolling the layer to form a multilayer device having channels.
28. (Currently amended) A method of making a multilayer device comprising the steps of:
- (a) obtaining layers of a multilayer device, comprising:
 - (i) at least a first layer comprised of a polymer scaffold ~~suitable~~ having a pattern of microchannels therein, and
 - (A) wherein the pattern of microchannels are suitable for the attachment and culturing of animal cells within the channels,
 - (B) wherein the pattern of microchannels are connected for the circulation of fluid through the layer; and
 - (C) wherein the first layer is formed by forming a first mold from a substrate material using a photoresist processing technique that includes creating a microfluidic circuit pattern on a substrate by using a manufacturing process coating the substrate material with a photoresist, and forming a pattern in the photoresist, and then casting an elastomer on the mold such that transferring the microfluidic circuit pattern in the photoresist is transferred to the an elastomer which acts as a second mold for the first layer polymer scaffold, and
 - (ii) at least a second layer, wherein the second layer is comprised of a polymer scaffold for attachment and culturing of animal cells; and
 - (b) fastening the at least a first layer and the at least a second layer together.

29. (Previously presented) The method of claim 28, wherein the animal cells are selected from the group consisting of endothelial cells, parenchymal cells, bone marrow cells, osteoblasts, mesenchymal stem cells, satellite cells, and fibroblasts.

30. (Previously presented) The method of claim 28, further comprising the step of (c) seeding animal cells to into the channels.

31. (Previously presented) The method of claim 28, wherein the animal cells are endothelial cells.

32. (Currently amended) A method of implanting a bioartificial organ into a recipient, comprising:

- (a) obtaining a multilayer device, comprising:
 - (i) at least a first layer comprised of a polymer scaffold having a pattern of microchannels therein, and
 - (A) wherein the microchannels are suitable for the attachment and culturing of animal cells within the channels,
 - (B) wherein the microchannels are connected for the circulation of fluid through the layer, and
 - (C) wherein the first layer is formed by forming a mold from a semiconductor substrate material using a photoresist processing technique that includes coating the semiconductor substrate material with a light sensitive photoresist, and forming a pattern in the photoresist by exposing the photoresist to short-wavelength light ~~polymer scaffold is molded by optically through a semi-transparent mask to create a microfluidic pattern in the photoresist~~ a light sensitive material on a substrate which acts as a mold, and then casting the first layer polymer scaffold on the mold, and removing the first layer polymer scaffold from the mold; and
 - (ii) at least a second layer wherein the second layer is comprised of a polymer scaffold, the layers of the multilayer device being fastened together; and
- (b) implanting the multilayer device into the recipient, wherein the implanted multilayer device is a bioartificial organ.

33. (Cancelled) An image reversal method for forming a scaffold having a material having a pattern of microchannels therein, comprising

- (a) selecting a mold having a complex pattern of microchannels on the mold in a reverse image of the channels; and
- (b) replica molding the pattern from the mold to a material suitable for attachment and culturing of animal cells;

wherein the replica molding forms a scaffold having a complex pattern of microchannels therein.

34. (Cancelled) A multilayer device, comprising:

- (a) multiple layers of tissue;
- (b) multiple layers of material suitable for attachment and growth of tissue and having a pattern of microchannels in the material;
- (b) vasculature within the tissue or on the material; and
- (c) connections for flow into and out of the vasculature.

35. (Cancelled) The multilayer device of claim 34, wherein the vasculature comprises endothelial cells.

36. (Cancelled) The multilayer device of claim 35, further comprising cells selected from the group consisting of parenchymal cells, cells forming cartilage or bone, muscle cells, and nerve cells.

37. (Cancelled) The multilayer device of claim 36, wherein the parenchymal cells are derived from organs selected from the group consisting of heart, liver, pancreas, intestine, kidney, reproductive tissues and lung.

38. (Currently amended) The multilayer device of claim 10 or 18, wherein the pattern in the first layer comprises microchannels that are about 30-200 microns in width ~~diameter~~.

39. (Previously presented) The multilayer device of claim 1, wherein the substrate is selected from the group consisting of silicon, ceramic, and glass.

40. (Previously presented) The multilayer device of claim 1, wherein the second layer is formed by forming a mold from a substrate selected from the group consisting of silicon, ceramic, and glass using a photoresist processing technique, and casting the second layer on the respective mold, wherein the photoresist processing technique for the second layer includes coating the substrate with a photoresist, forming a pattern in the photoresist to form a second mold and casting the second layer on the second mold.

41. (Previously presented) The multilayer device of claim 40, wherein the molds for the first and second layers are the same.
42. (Previously presented) The multilayer device of claim 40, wherein the molds for the first and second layers are different.
43. (Previously presented) The multilayer device of claim 1, wherein the polymer scaffold is selected from a material consisting of a biocompatible material, a biodegradable material, a porous material, a non-porous material and combinations thereof.
44. (Previously presented) The multilayer device of claim 1, wherein the microchannels are a branched pattern.
45. (Currently amended) The multilayer device of claim 1, wherein the microchannels have a height and width of that are about 30-200 microns.
46. (Previously presented) The multilayer device of claim 1, wherein the photoresist in combination with the substrate material form the mold.
47. (Currently Amended) The multilayer device of claim 1, wherein forming the mold includes is-formed by etching the substrate.
48. (Currently Amended) The multilayer device of claim 1, wherein forming the mold includes is-formed by etching the substrate in a pattern exposed through the photoresist and stripping the photoresist therefrom.
49. (Previously presented) The method of claim 25, wherein the lumens are substantially rectangular.
50. (Cancelled).
51. (Currently Amended) The method of claim 28, wherein the first layer polymer scaffold is compression molded on the mold.
52. (Previously presented) The method of claim 28, wherein the polymer scaffold is about 200 microns thick.
53. (Previously presented) The method of claim 28, wherein the microchannels are about 2 microns in width.
54. (Previously presented) The method of claim 28, further comprising the steps of implanting the multilayer device and seeding animal cells onto at least the first layer.
55. (Currently Amended) The method of claim ~~32~~8, wherein the elastomer is a polydimethylsiloxane elastomer.

56. (Previously presented) The method of claim 32, wherein the multilayer device is seeded with animal cells.

57. (Previously presented) The method of claim 56, wherein the multilayer device is seeded with animal cells after implanting.

58. (Currently amended) A multilayer device for use in tissue engineering, comprising:

- (a) at least a first layer comprised of a polymer scaffold having a pattern of microchannels therein and
 - (i) wherein the microchannels are suitable for the attachment and culturing of animal cells within the microchannels,
 - (ii) wherein the microchannels are connected for the circulation of fluid through the first layer, and
 - (iii) wherein the at least a first layer is fabricated by forming a mold from a semiconductor substrate material using a photoresist processing technique that includes ~~semiconductor manufacturing process~~ coating the semiconductor substrate material with a photoresist, and forming a pattern in the photoresist, and then casting the at least a first layer on the mold; and
- (b) at least a second layer comprised of a polymer scaffold,

wherein the first and second layers are joined or fastened together.

59. (Previously presented) The multilayer device of claim 1, wherein the photoresist processing technique further includes the step of etching the pattern into the substrate.

60. (Currently Amended) A method as recited in Claim 32, further comprising the step of etching the ~~microfluidic~~ pattern of microchannels in the substrate.